IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant:	CLEARY, et al.
Serial No.:	10/529,622
Filed:	March 30, 2005
For:	Highly Purified Amphotericin B
Group Art Unit:	1623
Examiner:	PRESELEV, Elli
Attorney's Docket No.	11636N/020724
Customer No.	32885

DECLARATION UNDER 37 C.F.R. § 1.132

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

I, John D. Cleary, Ph.D., declare:

1. I have significant experience in the field of pharmacology. My current position is Professor and Vice Chairman of Research, Schools of Medicine and Pharmacy; and Anti-mycotic Program Director, Mycotic Research Center; University of Mississippi Medical Center.

I consider myself to be one of ordinary skill in the art.

2. I have read and understood the Office Action mailed November 28, 2007, in connection with the above-identified application. Furthermore, I have read and understand the patents cited therein.

3. I, along with co-inventors Chapman and Kramer, am a co-author of High Purity Amphotericin B, *J. Antimicro Chemother* 2007 60(6): 1331-1340. A copy of the article is attached.

4. The article summarizes data related to compositions of the present invention, and shows superior and unexpected results of the present invention when compared to compositions of the prior art.

5. Highly purified amphotericin B [AmB]of the present invention was first isolated from a commercially generic AmB formulation by semi-preparative reverse phase high-pressure liquid chromatography and then its effects were compared *in vitro* and *in vivo* with those commercial formulations.

6. We found that general toxicity to highly purified AmB *in vivo*, as indicated by its apparent LD_{50} and survival of *Candida*-infected mice, was roughly 2-fold less than was toxicity to generic or lipid-complexed AmB.

Likewise, highly purified AmB decreased mean glomerular filtration rate about half as much as did a 10-fold greater dose of Pharma-Tek AmB. This result is superior and unexpected.

7. For this project, the nominal purity of the active ingredient was defined to be 95% AmB and no greater than 5% or less impurities. In practice, the apparent purity of the active ingredient varied between preparations from 96-99%, and corresponding impurity product percentages ranged from 4 to 1%.

8. The level of purity of the commercially available non-lipid AmB formulations such as Pharma-Tek® AmB was about 89%.

9. With respect to renal toxicity, the data indicated that renal toxicity associated with AmB formulations varied between agents. The final doses for the commercial comparison were 2 mg/kg for Pharma-Tek® AmB and 20 mg/kg for both the AmB lipid formulations and compositions of the present invention, each administered for one hour. When data are viewed as percent control, one can easily notice the differences in renal function (GFR) of animals treated with compositions of the present invention (7.9% decrease) or liposomal AmB (16.4% decrease) compared to the renal function of animals

treated with Pharma-Tek generic AmB (12.3% decrease). This result is superior and unexpected.

10. With respect to mortality, the compositions of the present invention appeared to have the greatest efficacy. Mortality in infected mice treated with compositions of the present invention was only 20%, whereas mortality in animals treated with either Pharma-Tek® AmB or AmBLC was 40%. Again, the compositions of the present invention were given at a 10-fold greater dose.

11. Drug-associated renal dysfunction is among the most clinically important AmB side effects. Pharmacoeconomic assessments of the cost of using AmB have yielded frightening results. An estimated 30% of patients treated for systemic fungal infections will experience severe renal dysfunction (renal failure) and will require, on average, an additional 8.2 days of hospitalization along with a secondary 2- to 2.7-fold increased risk of death. The cost of AmB-induced events was $29,823 per case. The use of lipid-based formulations of AmB, secondary to their lower risk for nephrotoxicity, is replacing conventional AmB therapy for treatment of systemic fungal infection except in many HIV-infected and pediatric patients. Yet, the cost of comparable therapy is considerably greater for the lipid formulation; daily

cost for AmB averages $25, whereas that for lipid-formulated AmB ranges between $450 and $1850. Assuming a 14-day course of therapy, a patient will pay an average of $7000 more for a lipid-based, albeit safer, AmB product. Thus, the present invention can provide a significant improvement over what is currently available. Given the superior and unexpected improvements of the present invention over previous attempts to make AmB treatment safer, the present invention can potentially increase the number of treatment candidates, and allow for increased dosages with reduced side effects.

12. The undersigned declares further that all statements made herein of his knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under section 1001 of Title 18 of United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Signed this 29 day of April, 2008.

John D. Cleary, Pharm.D.